PCT/EP2004/011619

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Dated 8 October 2004

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Request for grant of a patent

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The Patent Office
Cardiff Road
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1. Your reference — MG/PMS/PB60543P

2. Patent application number
(The Patent Office will fill in his part) — 0324159.3 — 15 OCT 2003

3. Full name, address and postcode of the or of each applicant *(underline all surnames)*

Patents ADP number *(if you know it)*

If the applicant is a corporate body, give the country/state of its incorporation

Glaxo Group Limited
Glaxo Wellcome House, Berkeley Avenue,
Greenford, Middlesex UB6 0NN, Great Britain

473587003

United Kingdom

4. Title of the invention — Novel Compounds

5. Name of your agent *(if you have one)*

"Address for service" in the United Kingdom to which all correspondence should be sent *(including the postcode)*

Patents ADP number *(if you know it)*

Corporate Intellectual Property

GlaxoSmithKline
Corporate Intellectual Property (CN9 25.1)
980 Great West Road
BRENTFORD
Middlesex TW8 9GS

8072555006

6. If you are declaring priority from one or more earlier patent applications, give the country and the date of filing of the or each of these earlier applications and *(if you know it)* the or each application number

Country	Priority application number *(if you know it)*	Date of filing *(day / month / year)*

7. If this application is divided or otherwise derived from an earlier UK application, give the number and the filing date of the earlier application

Number of earlier application	Date of filing *(day / month / year)*

8. Is a statement of inventorship and of right to grant of a patent required in support of this request? *(Answer yes if:*

a) any applicant named in part 3 is not an inventor, or
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Patents Form 1/77

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Continuation sheets of this form	0
Description	15
Claim(s)	2
Abstract	0
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Priority Documents	
Translations of priority documents	
Statement of inventorship and right to grant of a patent *(Patents Form 7/77)*	
Request for preliminary examination and search *(Patents Form 9/77)*	
Request for substantive examination *(Patents Form 10/77)*	
Any other documents *(please specify)*	

11. We request the grant of a patent on the basis of this application
Signature M Gibson Date 15-Oct-03

12. Name and daytime telephone number of person to contact in the United Kingdom — M Gibson 01279 644841

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PB60543P

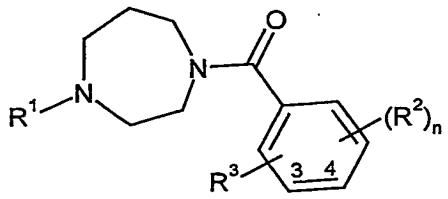
NOVEL COMPOUNDS

The present invention relates to novel diazepanyl derivatives having pharmacological activity, processes for their preparation, to compositions containing them and to their use in the treatment of neurological and psychiatric disorders.

WO 03/00480 (Novo Nordisk A/S and Boehringer Ingleheim International GMBH) describes a series of substituted piperazines and diazepanes as H3 antagonists. WO 02/08221 (Neurogen Corporation) describes a series of substituted piperazines and diazepanes as capsaicin receptor antagonists which are claimed to be useful in the treatment of neuropathic pain. WO 98/37077 and WO 99/42107 (Zymogenetics Inc) both describe a series of substituted heterocyclic derivatives which are claimed to act as calcitonin mimics to enhance bone formation.

The histamine H3 receptor is predominantly expressed in the mammalian central nervous system (CNS), with minimal expression in peripheral tissues except on some sympathetic nerves (Leurs *et al.*, (1998), Trends Pharmacol. Sci. **19**, 177-183). Activation of H3 receptors by selective agonists or histamine results in the inhibition of neurotransmitter release from a variety of different nerve populations, including histaminergic and cholinergic neurons (Schlicker *et al.*, (1994), Fundam. Clin. Pharmacol. **8**, 128-137). Additionally, *in vitro* and *in vivo* studies have shown that H3 antagonists can facilitate neurotransmitter release in brain areas such as the cerebral cortex and hippocampus, relevant to cognition (Onodera *et al.*, (1998), In: The Histamine H3 receptor, ed Leurs and Timmerman, pp255-267, Elsevier Science B.V.). Moreover, a number of reports in the literature have demonstrated the cognitive enhancing properties of H3 antagonists (e.g. thioperamide, clobenpropit, ciproxifan and GT-2331) in rodent models including the five choice task, object recognition, elevated plus maze, acquisition of novel task and passive avoidance (Giovanni *et al.*, (1999), Behav. Brain Res. **104**, 147-155). These data suggest that novel H3 antagonists and/or inverse agonists such as the current series could be useful for the treatment of cognitive impairments in neurological diseases such as Alzheimer's disease and related neurodegenerative disorders.

The present invention provides, in a first aspect, a compound of formula (I) or a pharmaceutically acceptable salt thereof:



(I)

wherein:

R^1 represents branched C_{3-6} alkyl, C_{3-5} cycloalkyl or -C_{1-4} alkylC_{3-4} cycloalkyl;
R^2 represents halogen, C_{1-6} alkyl, C_{1-6} alkoxy, cyano, amino or trifluoromethyl;
n represents 0, 1 or 2;
R^3 represents –X-aryl, -X-heteroaryl, -X-heterocyclyl, -X-aryl-aryl, -X-aryl-heteroaryl, -X-aryl-heterocyclyl, -X-heteroaryl-aryl, -X-heteroaryl-heteroaryl, -X-heteroaryl-heterocyclyl, -X-heterocyclyl-aryl, -X-heterocyclyl-heteroaryl or –X-heterocyclyl-heterocyclyl;
such that when R^3 represents –X-piperidinyl, -X-piperidinyl-aryl, -X-piperidinyl-heteroaryl or –X-piperidinyl-heterocyclyl said piperidinyl group is attached to X via a nitrogen atom;
wherein R^3 is attached to the phenyl group of formula (I) at the 3 or 4 position;
X represents a bond, O, CO, SO_2, CH_2O, OCH_2 or C_{1-6} alkyl;
wherein said aryl, heteroaryl or heterocyclyl groups of R^3 may be optionally substituted by one or more (eg. 1, 2 or 3) halogen, hydroxy, cyano, nitro, oxo, haloC_{1-6} alkyl, polyhaloC_{1-6} alkyl, haloC_{1-6} alkoxy, polyhaloC_{1-6} alkoxy, C_{1-6} alkyl, C_{1-6} alkoxy, arylC_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkoxyC_{1-6} alkyl, C_{3-7} cycloalkylC_{1-6} alkoxy, C_{3-7} cycloalkylcarbonyl, -COC_{1-6} alkyl, C_{1-6} alkoxycarbonyl, arylC_{1-6} alkyl, heteroarylC_{1-6} alkyl, heterocyclylC_{1-6} alkyl, C_{1-6} alkylsulfonyl, C_{1-6} alkylsulfinyl, C_{1-6} alkylsulfonyloxy, C_{1-6} alkylsulfonylC_{1-6} alkyl, arylsulfonyl, arylsulfonyloxy, arylsulfonylC_{1-6} alkyl, aryloxy, -CO-aryl, -CO-heterocyclyl, -CO-heteroaryl, C_{1-6} alkylsulfonamidoC_{1-6} alkyl, C_{1-6} alkylamidoC_{1-6} alkyl, arylsulfonamido, arylaminosulfonyl, arylsulfonamidoC_{1-6} alkyl, arylcarboxamidoC_{1-6} alkyl, aroylC_{1-6} alkyl, arylC_{1-6} alkanoyl, or a group $NR^{15}R^{16}$, -NR^{15}CO-aryl, -NR^{15}CO-heterocyclyl, -NR^{15}CO-heteroaryl, -CO$NR^{15}R^{16}$, -$NR^{15}COR^{16}$, -$NR^{15}SO_2R^{16}$ or -$SO_2NR^{15}R^{16}$ groups, wherein R^{15} and R^{16} independently represent hydrogen or C_{1-6} alkyl;
or solvates thereof.

Alkyl groups, whether alone or as part of another group, may be straight chain or branched and the groups alkoxy and alkanoyl shall be interpreted similarly. The term 'halogen' is used herein to describe, unless otherwise stated, a group selected from fluorine, chlorine, bromine or iodine and the term 'polyhalo' is used herein to refer to a moiety containing more than one (eg. 2-5) of said halogen atoms. Branched alkyl refers to an alkyl chain which is branched at the first carbon atom (eg. isopropyl, isobutyl or tertiary butyl).

The term "aryl" includes single and fused rings wherein at least one ring is aromatic, for example, phenyl, naphthyl and tetrahydronaphthalenyl.

The term "heterocyclyl" is intended to mean a 4-7 membered monocyclic saturated or partially unsaturated aliphatic ring containing 1 to 3 heteroatoms selected from oxygen or nitrogen. Suitable examples of such monocyclic rings include pyrrolidinyl, piperidinyl, piperazinyl, morpholinyl, 1,3-dioxolane, diazepanyl and azepanyl.

The term "heteroaryl" is intended to mean a 5-7 membered monocyclic aromatic or a fused 8-11 membered bicyclic aromatic ring containing 1 to 3 heteroatoms selected from

oxygen, nitrogen and sulphur. Suitable examples of such monocyclic aromatic rings include thienyl, furyl, pyrrolyl, triazolyl, imidazolyl, oxazolyl, thiazolyl, oxadiazolyl, isothiazolyl, isoxazolyl, thiadiazolyl, pyrazolyl, pyrimidyl, pyridazinyl, pyrazinyl and pyridyl. Suitable examples of such fused aromatic rings include benzofused aromatic rings such as quinolinyl, isoquinolinyl, quinazolinyl, quinoxalinyl, cinnolinyl, naphthyridinyl, indolyl, indazolyl, pyrrolopyridinyl, benzofuranyl, benzothienyl, benzimidazolyl, benzoxazolyl, benzisoxazolyl, benzothiazolyl, benzisothiazolyl, benzoxadiazolyl, benzothiadiazolyl and the like.

Preferably, R^1 represents branched C_{3-6} alkyl (eg. isopropyl) or C_{3-5} cycloalkyl (eg. cyclopropyl or cyclobutyl), more preferably cyclobutyl.

Preferably, n represents 0.

Preferably, R^3 represents

–X-aryl (eg. –phenyl, -CO-phenyl, -O-phenyl or $–OCH_2$-phenyl) optionally substituted by one or more cyano or $-COC_{1-6}$ alkyl (eg. –COMe) groups;

-X-heteroaryl (eg. –tetrazolyl);

-X-heteroaryl-aryl (eg. –thiazolyl-phenyl) optionally substituted by one or more halogen (eg. fluorine) atoms; or

-X-heterocyclyl (eg. thiomorpholinyl or o-tetrahydro-2H-pyran-4-yl) optionally substituted by one or more oxo groups.

More preferably, R^3 represents –X-aryl (eg. –phenyl, -CO-phenyl, -O-phenyl or $–OCH_2$-phenyl) optionally substituted by one or more cyano or $-COC_{1-6}$ alkyl (eg. –COMe) groups.

Preferably, X represents a bond, CO, O or OCH_2 more preferably a bond.

Preferably, R^3 is attached to the phenyl group of formula (I) at the 4 position.

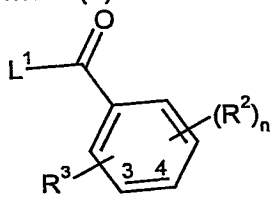
Preferred compounds according to the invention include examples E1-E10 as shown below, or a pharmaceutically acceptable salt thereof.

Compounds of formula (I) may form acid addition salts with acids, such as conventional pharmaceutically acceptable acids, for example maleic, hydrochloric, hydrobromic, phosphoric, acetic, fumaric, salicylic, sulphate, citric, lactic, mandelic, tartaric and methanesulphonic. Salts, solvates and hydrates of histamine H3 receptor antagonists or inverse agonists therefore form an aspect of the invention.

Certain compounds of formula (I) are capable of existing in stereoisomeric forms. It will be understood that the invention encompasses all geometric and optical isomers of these compounds and the mixtures thereof including racemates. Tautomers also form an aspect of the invention.

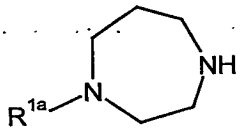
The present invention also provides a process for the preparation of a compound of formula (I) or a pharmaceutically acceptable salt thereof, which process comprises:

(a) reacting a compound of formula (II)



(II)

wherein R^2 and R^3 are as defined above and L^1 represents OH or a suitable leaving group, such as a halogen atom (eg. chlorine), with a compound of formula (III)



(III)

wherein R^{1a} is as defined above for R^1 or is a group convertible to R^1;

(b) deprotecting a compound of formula (I) which is protected; and optionally thereafter

(c) interconversion to other compounds of formula (I).

Process (a) typically comprises activation of the compound of formula (II) wherein L^1 represents OH with a coupling reagent such as 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (EDC) in the presence of 1-hydroxybenzotriazole (HOBT) in a suitable solvent such as dichloromethane followed by reaction with the compound of formula (III).

Process (a) may also involve halogenation of the compound of formula (II) wherein L^1 represents OH with a suitable halogenating agent (eg. thionyl chloride or oxalyl chloride) followed by reaction with the compound of formula (III) in the presence of a suitable base such as triethylamine or a solid supported base such as diethylaminomethylpolystyrene in a suitable solvent such as dichloromethane.

In process (b), examples of protecting groups and the means for their removal can be found in T. W. Greene 'Protective Groups in Organic Synthesis' (J. Wiley and Sons, 1991). Suitable amine protecting groups include sulphonyl (e.g. tosyl), acyl (e.g. acetyl, 2',2',2'-trichloroethoxycarbonyl, benzyloxycarbonyl or t-butoxycarbonyl) and arylalkyl (e.g. benzyl), which may be removed by hydrolysis (e.g. using an acid such as hydrochloric acid) or reductively (e.g. hydrogenolysis of a benzyl group or reductive

removal of a 2',2',2'-trichloroethoxycarbonyl group using zinc in acetic acid) as appropriate. Other suitable amine protecting groups include trifluoroacetyl (-$COCF_3$) which may be removed by base catalysed hydrolysis or a solid phase resin bound benzyl group, such as a Merrifield resin bound 2,6-dimethoxybenzyl group (Ellman linker), which may be removed by acid catalysed hydrolysis, for example with trifluoroacetic acid.

Process (c) may be performed using conventional interconversion procedures such as epimerisation, oxidation, reduction, alkylation, nucleophilic or electrophilic aromatic substitution, ester hydrolysis or amide bond formation.

Compounds of formula (II) and (III) are either known in the literature or can be prepared by analogous methods.

Compounds of formula (I) and their pharmaceutically acceptable salts have affinity for and are antagonists and/or inverse agonists of the histamine H3 receptor and are believed to be of potential use in the treatment of neurological diseases including Alzheimer's disease, dementia, age-related memory dysfunction, mild cognitive impairment, cognitive deficit, epilepsy, neuropathic pain, inflammatory pain, migraine, Parkinson's disease, multiple sclerosis, stroke and sleep disorders including narcolepsy; psychiatric disorders including schizophrenia (particularly cognitive deficit of schizophrenia), attention deficit hypereactivity disorder, depression and addiction; and other diseases including obesity, asthma, allergic rhinitis, nasal congestion, chronic obstructive pulmonary disease and gastro-intestinal disorders.

Thus the invention also provides a compound of formula (I) or a pharmaceutically acceptable salt thereof, for use as a therapeutic substance in the treatment or prophylaxis of the above disorders, in particular cognitive impairments in diseases such as Alzheimer's disease and related neurodegenerative disorders.

The invention further provides a method of treatment or prophylaxis of the above disorders, in mammals including humans, which comprises administering to the sufferer a therapeutically effective amount of a compound of formula (I) or a pharmaceutically acceptable salt thereof.

In another aspect, the invention provides the use of a compound of formula (I) or a pharmaceutically acceptable salt thereof in the manufacture of a medicament for use in the treatment of the above disorders.

When used in therapy, the compounds of formula (I) are usually formulated in a standard pharmaceutical composition. Such compositions can be prepared using standard procedures.

Thus, the present invention further provides a pharmaceutical composition for use in the treatment of the above disorders which comprises the compound of formula (I) or a pharmaceutically acceptable salt thereof and a pharmaceutically acceptable carrier.

The present invention further provides a pharmaceutical composition which comprises the compound of formula (I) or a pharmaceutically acceptable salt thereof and a pharmaceutically acceptable carrier.

Compounds of formula (I) may be used in combination with other therapeutic agents, for example histamine H1 antagonists or medicaments claimed to be useful as either disease modifying or symptomatic treatments of Alzheimer's disease. Suitable examples of such other therapeutic agents may be agents known to modify cholinergic transmission such as 5-HT_6 antagonists, M1 muscarinic agonists, M2 muscarinic antagonists or acetylcholinesterase inhibitors. When the compounds are used in combination with other therapeutic agents, the compounds may be administered either sequentially or simultaneously by any convenient route.

The invention thus provides, in a further aspect, a combination comprising a compound of formula (I) or a pharmaceutically acceptable derivative thereof together with a further therapeutic agent or agents.

The combinations referred to above may conveniently be presented for use in the form of a pharmaceutical formulation and thus pharmaceutical formulations comprising a combination as defined above together with a pharmaceutically acceptable carrier or excipient comprise a further aspect of the invention. The individual components of such combinations may be administered either sequentially or simultaneously in separate or combined pharmaceutical formulations.

When a compound of formula (I) or a pharmaceutically acceptable derivative thereof is used in combination with a second therapeutic agent active against the same disease state the dose of each compound may differ from that when the compound is used alone. Appropriate doses will be readily appreciated by those skilled in the art.

A pharmaceutical composition of the invention, which may be prepared by admixture, suitably at ambient temperature and atmospheric pressure, is usually adapted for oral, parenteral or rectal administration and, as such, may be in the form of tablets, capsules, oral liquid preparations, powders, granules, lozenges, reconstitutable powders, injectable or infusible solutions or suspensions or suppositories. Orally administrable compositions are generally preferred.

Tablets and capsules for oral administration may be in unit dose form, and may contain conventional excipients, such as binding agents, fillers, tabletting lubricants, disintegrants and acceptable wetting agents. The tablets may be coated according to methods well known in normal pharmaceutical practice.

Oral liquid preparations may be in the form of, for example, aqueous or oily suspension, solutions, emulsions, syrups or elixirs, or may be in the form of a dry product for reconstitution with water or other suitable vehicle before use. Such liquid preparations may contain conventional additives such as suspending agents, emulsifying agents, non-aqueous vehicles (which may include edible oils), preservatives, and, if desired, conventional flavourings or colorants.

For parenteral administration, fluid unit dosage forms are prepared utilising a compound of the invention or pharmaceutically acceptable salt thereof and a sterile vehicle. The compound, depending on the vehicle and concentration used, can be either suspended or dissolved in the vehicle. In preparing solutions, the compound can be dissolved for injection and filter sterilised before filling into a suitable vial or ampoule and sealing. Advantageously, adjuvants such as a local anaesthetic, preservatives and buffering agents are dissolved in the vehicle. To enhance the stability, the composition can be frozen after filling into the vial and the water removed under vacuum. Parenteral suspensions are prepared in substantially the same manner, except that the compound is suspended in the vehicle instead of being dissolved, and sterilisation cannot be accomplished by filtration. The compound can be sterilised by exposure to ethylene oxide before suspension in a sterile vehicle. Advantageously, a surfactant or wetting agent is included in the composition to facilitate uniform distribution of the compound.

The composition may contain from 0.1% to 99% by weight, preferably from 10 to 60% by weight, of the active material, depending on the method of administration. The dose of the compound used in the treatment of the aforementioned disorders will vary in the usual way with the seriousness of the disorders, the weight of the sufferer, and other similar factors. However, as a general guide suitable unit doses may be 0.05 to 1000 mg, more suitably 1.0 to 200 mg, and such unit doses may be administered more than once a day, for example two or three a day. Such therapy may extend for a number of weeks or months.

The following Descriptions and Examples illustrate the preparation of compounds of the invention.

Description 1

1-*tert*-Butyl-4-(isopropyl)-hexahydro-1*H*-1,4-diazepine-1-carboxylate (D1)

tert-Butyl-hexahydro-1*H*-1,4-diazepine-1-carboxylate (10.0g) was dissolved in DCM (200ml). Acetone (7.33ml) was added and the reaction was left to stir for 5min. Sodium

triacetoxyborohydride (21.0g) was then added and the reaction was stirred at rt for 16h. The reaction mixture was washed with saturated potassium carbonate solution (2 x 200ml). The organic layer was dried (magnesium sulphate) and evaporated to give the title compound (D1) as a clear oil (11.0g).

Description 2

1-(Isopropyl)-hexahydro-1*H*-1,4-diazepine dihydrochloride (D2)

1-*tert*-Butyl-4-(isopropyl)-hexahydro-1*H*-1,4-diazepine-1-carboxylate (D1) (11.0g) was dissolved in methanol (200ml) and 4N HCl in dioxan (100ml) was added. The reaction was stirred at rt for 2h and then evaporated to give the title compound (D2) as a white solid (9.6g). ^{1}H NMR δ ($CDCl_3$): 11.35 (1H, s), 10.22 (1H, s), 9.72 (1H, s), 4.15-3.52 (9H, m), 2.83-2.40 (2H, m), 1.47 (6H, d, J=6.24 Hz).

Description 3

1-*tert*-Butyl-4-(cyclobutyl)-hexahydro-1*H*-1,4-diazepine-1-carboxylate (D3)

tert-Butyl-hexahydro-1*H*-1,4-diazepine-1-carboxylate (10.0g) was dissolved in DCM (300ml). Cyclobutanone (7.5ml) was added and the reaction was left to stir for 5 min. Sodium triacetoxyborohydride (21.1g) was then added and the reaction was stirred at rt for 16h. The reaction mixture was washed with saturated potassium carbonate solution (2 x 200ml). The organic layer was dried (magnesium sulphate) and evaporated to give the title compound (D3) as a clear oil (11.3g).

Description 4

1-(Cyclobutyl)hexahydro-1*H*-1,4-diazepine dihydrochloride (D4)

1-*tert*-Butyl-4-(cyclobutyl)-hexahydro-1*H*-1,4-diazepine-1-carboxylate (D3) (11.3g) was dissolved in methanol (200ml) and 4N HCl in dioxan (100ml) was added. The reaction was stirred at rt for 3h and then co-evaporated from toluene (3 x 50ml) to give the title compound (D4) as a white solid (9.8g). ^{1}H NMR δ (DMSO-d6): 11.95 (1H, s), 9.55 (1H, s), 9.64 (1H, s), 3.78-3.08 (9H, m), 2.51-2.07 (6H, m), 1.80-1.51 (2H, m).

Description 5

Ethyl 4-(tetrahydro-2*H*-pyran-4-yloxy)benzoate (D5)

An ice-cold solution of ethyl 4-hydroxybenzoate (0.82g), 4-hydroxy-tetrahydro-2H-pyran (0.5g) and triphenylphosphine in THF (50ml) was treated dropwise with diisopropyl azodicarboxylate (1.69ml). After 15min the cooling bath was removed and the reaction stood overnight at rt. The mixture was evaporated, redissolved in toluene and successively washed with 2N sodium hydroxide (2x20ml), water (2x20ml) and brine (20ml). After drying (magnesium sulfate) the solution was loaded directly on to a silica flash column (step gradient 10-30% EtOAc in light petroleum 40-60) to give the title compound (D5) (0.75g). ^{1}H NMR δ ($CDCl_3$): 7.98 (2H, d, J=8.5Hz), 6.91 (2H, d, J=8.5Hz), 4.60 (1H, m), 4.35 (2H, q, J=9.8Hz), 3.98 (2H, m), 3.57 (2H, m), 2.05 (2H, m), 1.80 (2H, m), 1.38 (3H, t, J=9.8Hz).

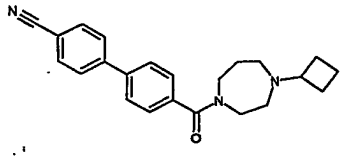
Description 6

4-(Tetrahydro-2*H*-pyran-4-yloxy)benzoic acid (D6)

A solution of ethyl 4-(tetrahydro-2*H*-pyran-4-yloxy)benzoate (D5) (0.73g) in EtOH (10ml) was treated with 1M NaOH (5.84ml) and the mixture stirred at 60°C for 5h. The solution was cooled to rt and the EtOH was evaporated. The aqueous was washed with DCM (2x10ml) and acidified. The solid was filtered off, washed with water and dried to give the title compound (D6) (0.55g). MS electrospray (-ion) 221 (M-H). ^{1}H NMR δ (DMSO-d6): 7.87 (2H, d, J=8.5Hz), 7.05 (2H, d, J=8.5Hz), 4.69 (1H, m), 3.85 (2H, m), 3.50 (2H, m), 1.98 (2H, m), 1.59 (2H, m).

Example 1

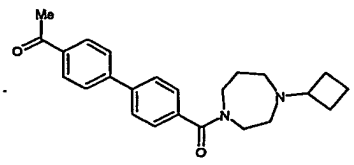
4'-[(4-Cyclobutylhexahydro-1*H*-1,4-diazepin-1-yl)carbonyl]-4-biphenylcarbonitrile hydrochloride (E1)



1-(Cyclobutyl)-hexahydro-1*H*-1,4-diazepine dihydrochloride (D4) (0.15g) was stirred with diethylaminomethyl polystyrene (1.0g), HOBT (0.045g), 4'-cyano-4-biphenylcarboxylic acid (0.16g) in DCM (5ml). EDC (0.16g) was then added and the reaction was stirred at rt for 16h. The polymer supported base was filtered off and the filtrate was diluted with DCM (10ml) and washed with saturated sodium hydrogen carbonate (2 x 15ml). The organic layer was then loaded directly onto a silica column eluting with 0-10% MeOH (containing 10% 0.880 ammonia solution)/DCM. The isolated free base product was dissolved in DCM (5ml) and treated with excess1N HCl/diethyl ether solution (1ml) and stirred for 10min. The mixture was evaporated (co-evaporated with acetone 2 x 10ml), triturated with acetone, then dried at 50°C under high vacuum for 16h to yield the title compound (E1) as a pale solid (0.119g). MS electrospray (+ion) 360 (MH^+). ^{1}H NMR δ (DMSO-d6): 10.60 (1H, s), 7.97 (4H, m), 7.86 (2H, d, J=8.4Hz), 7.60 (2H, d, J= 7.6Hz), 4.18 (1H, m), 3.89-3.37 (6H, m), 3.10 (2H, m), 2.40-1.59 (8H, m).

Example 2

1-{4'-[(4-Cyclobutylhexahydro-1*H*-1,4-diazepin-1-yl)carbonyl]-4-biphenylyl}ethanone hydrochloride (E2)

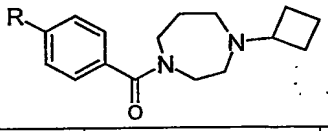


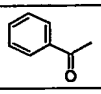
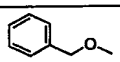
1-(Cyclobutyl)-hexahydro-1*H*-1,4-diazepine dihydrochloride (D4) (0.15g) was stirred with diethylaminomethyl polystyrene (1.0g), HOBT (0.045g) and 4'-acetyl-4-

biphenylcarboxylic acid (0.13g) in DCM (5ml). EDC (0.16g) was then added and the reaction stirred at rt for 16h. The polymer supported base was filtered off and the filtrate was diluted with DCM (10ml) and washed with saturated sodium hydrogen carbonate (2 x 15ml). The organic layer was loaded directly onto a silica column eluting with 0-10% MeOH (containing 10% 0.880 ammonia solution)/DCM. The isolated free base product was dissolved in DCM (5ml) and treated with excess1N HCl/diethyl ether solution (1ml) and stirred for 10min. The mixture was evaporated (co-evaporated with acetone 2 x 10ml), triturated with acetone, then dried at 50°C under high vacuum for 16h to yield the title compound (E2) as a pale solid (0.055g). MS electrospray (+ion) 377 (MH^+). ^1H NMR δ (DMSO-d6): 10.57 (1H, s), 9.07 (2H, d, J=6.4Hz), 7.88 (4H, m), 7.60 (2H, d, J= 7.6Hz), 4.15 (1H, m), 3.82-3.33 (6H, m), 3.02 (2H, m), 2.62 (3H, s), 2.41-1.62 (8H, m).

Examples 3-6 (E3-E6)

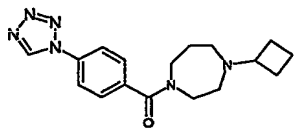
Examples 3 - 6 were prepared from 1-(cyclobutyl)-hexahydro-1*H*-1,4-diazepine dihydrochloride (D4) and the appropriate carboxylic acid, using the procedure described in Example 1 and displayed ^1H NMR and mass spectral data that were consistent with structure.



Example No	R	Mass Spectrum (ES^+)
E3		$[MH]^+$ 335
E4		$[MH]^+$ 363
E5		$[MH]^+$ 351
E6		$[MH]^+$ 365

Example 7

1-Cyclobutyl-4-{[4-tetrazol-1-yl)phenyl]carbonyl}hexahydro-1*H*-1,4-diazepine hydrochloride (E7)

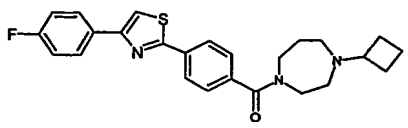


1-(Cyclobutyl)-hexahydro-1*H*-1,4-diazepine dihydrochloride (D4) (0.15g) was stirred with diethylaminomethyl polystyrene (1.0g), HOBT (0.045g) and 4-(tetrazol-1-yl)-benzoic acid (0.14g) in DCM (5ml). EDC (0.165g) was then added and the reaction was stirred at rt for 16h. The polymer supported base was filtered off and the filtrate was diluted with

DCM (10ml) and washed with saturated sodium hydrogen carbonate (2 x 15ml). The organic layer was then loaded directly onto a silica column eluting with 0-10% MeOH (containing 10% 0.880 ammonia solution)/DCM. The isolated free base product was dissolved in DCM (5ml) and treated with excess1N HCl/diethyl ether solution (1ml) and stirred for 10min. The mixture was evaporated (co-evaporated with acetone 2 x 10ml), triturated with acetone, then dried at 50°C under high vacuum for 16h to yield the title compound (E7) as a pale solid (0.096g). MS electrospray (+ion) 327 (MH^+). ^1H NMR δ (DMSO-d6): 11.11 (1H, s), 10.18 (1H, s), 8.02 (2H, d, J=8.4Hz), 7.76 (2H, d, J=8.0Hz), 4.17 (1H, m), 3.81-3.27 (6H, m), 3.11 (2H, m), 2.47-1.95 (6H, m), 1.80-1.59 (2H, m).

Example 8

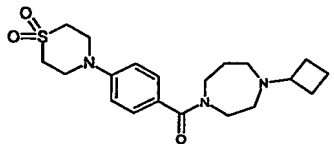
1-Cyclobutyl-4-({4-[4-(4-fluorophenyl)-1,3-thiazol-2-yl]phenyl}carbonyl) hexahydro-1*H*-1,4-diazepine hydrochloride (E8)



The title compound (E8) was prepared from 1-(cyclobutyl)-hexahydro-1*H*-1,4-diazepine dihydrochloride (D4) and 4-[4-(4-fluorophenyl)-1,3-thiazol-2-yl]benzoic acid using the procedure described in Example 7. MS electrospray (+ion) 437 (MH^+).

Example 9

1-Cyclobutyl-4-{[4-(1,1-dioxido-4-thiomorpholinyl)phenyl]carbonyl} hexahydro-1*H*-1,4-diazepine hydrochloride (E9)

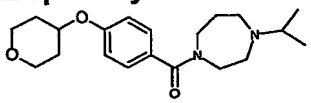


1-(Cyclobutyl)-hexahydro-1*H*-1,4-diazepine dihydrochloride (D4) (0.15g) was stirred with diethylaminomethyl polystyrene (1.0g), HOBT (0.045g), 4-(1,1-dioxido-4-thiomorpholinyl)benzoic acid (0.186g) in DCM (5ml). EDC (0.165g) was then added and the reaction was stirred at rt for 16h. The polymer supported base was filtered off and the filtrate was diluted with DCM (10ml) and washed with saturated sodium hydrogen carbonate (2 x 15ml). The organic layer was then loaded directly onto a silica column and eluted with 0-10% MeOH (containing 10% 0.880 ammonia solution)/DCM. The isolated free base product was dissolved in DCM (5ml) and treated with excess1N HCl/diethyl ether solution (1ml) and stirred for 10min. The mixture was evaporated (co-evaporated with acetone 2 x 10ml), triturated with acetone, then dried at 50°C under high vacuum for 16h to yield the title compound (E9) as a pale solid (0.086g). MS electrospray (+ion) 392 (MH^+). ^1H NMR δ (DMSO-d6): 10.5 (1H, s), 7.37 (2H, d,

J=8.4Hz), 7.07 (2H, d, J=8.8Hz), 4.18-3.24 (10H, m), 3.11 (4H, m), 3.10-2.85 (2H, m), 2.45-1.98 (7H, m), 1.80-2.54 (2H, m).

Example 10

1-(Isopropyl)-4-{[4-(tetrahydro-2*H*-pyran-4-yloxy)phenyl] carbonyl}hexahydro-1*H*-1,4-diazepine hydrochloride (E10)



A stirred suspension of 4-(tetrahydro-2*H*-pyran-4-yloxy)benzoic acid (D6) (222mg) in DCM (5ml) at rt was treated with oxalyl chloride (0.28ml) and 10% DMF in DCM (1 drop). After 1h the solution was evaporated and then re-evaporated from DCM (2x5ml). The acid chloride was redissolved in DCM (10ml) and treated with 1-(isopropyl)-hexahydro-1*H*-1,4-diazepine dihydrochloride (D2) (178mg) and diethylaminomethyl polystyrene (3.2mmol/g, 938mg). After stirring overnight the mixture was loaded directly on to a silica gel flash column [step gradient 6-10% MeOH (containing 10% 0.880 ammonia solution) in DCM]. Fractions containing the required product were evaporated, then redissolved in DCM and treated with excess 4M HCl in dioxan. Crystallisation from acetone afforded the title compound (E10) (225mg). MS electrospray (+ion) 347 (MH^+). ^{1}H NMR δ (DMSO-d6): 10.45 (1H, m), 7.41 (2H, d, J=8.5Hz), 7.02 (2H, d, J=8.5Hz), 4.63 (2H, m), 4.02 (1H, m), 3.02-3.93 (13H, m), 2.32 (1H, m), 1.96 (2H, m), 1.61 (2H, m), 1.27 (6H, d, J=6.5Hz).

Abbreviations

Boc	tert-butoxycarbonyl
EtOAc	ethyl acetate
h	hour
min	minutes
DCM	dichloromethane
MeOH	methanol
rt	room temperature
DMF	dimethylformamide
TFA	trifluoroacetic acid
HOBT	1-hydroxybenzotriazole
EDC	1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride

All publications, including but not limited to patents and patent applications, cited in this specification are herein incorporated by reference as if each individual publication were specifically and individually indicated to be incorporated by reference herein as though fully set forth.

Biological Data

A membrane preparation containing histamine H3 receptors may be prepared in accordance with the following procedures:

(i) Generation of histamine H3 cell line

DNA encoding the human histamine H3 gene (Huvar, A. *et al.* (1999) Mol. Pharmacol. **55(6)**, 1101-1107) was cloned into a holding vector, pCDNA3.1 TOPO (InVitrogen) and its cDNA was isolated from this vector by restriction digestion of plasmid DNA with the enzymes BamH1 and Not-1 and ligated into the inducible expression vector pGene (InVitrogen) digested with the same enzymes. The GeneSwitch™ system (a system where in transgene expression is switched off in the absence of an inducer and switched on in the presence of an inducer) was performed as described in US Patent nos: 5,364,791; 5,874,534; and 5,935,934. Ligated DNA was transformed into competent DH5α E. coli host bacterial cells and plated onto Luria Broth (LB) agar containing Zeocin™ (an antibiotic which allows the selection of cells expressing the sh ble gene which is present on pGene and pSwitch) at 50μg ml^{-1}. Colonies containing the re-ligated plasmid were identified by restriction analysis. DNA for transfection into mammalian cells was prepared from 250ml cultures of the host bacterium containing the pGeneH3 plasmid and isolated using a DNA preparation kit (Qiagen Midi-Prep) as per manufacturers guidelines (Qiagen).

CHO K1 cells previously transfected with the pSwitch regulatory plasmid (InVitrogen) were seeded at 2x10e6 cells per T75 flask in Complete Medium, containing Hams F12 (GIBCOBRL, Life Technologies) medium supplemented with 10% v/v dialysed foetal bovine serum, L-glutamine, and hygromycin (100μg ml^{-1}), 24 hours prior to use. Plasmid DNA was transfected into the cells using Lipofectamine plus according to the manufacturers guidelines (InVitrogen). 48 hours post transfection cells were placed into complete medium supplemented with 500μg ml^{-1} Zeocin™.

10-14 days post selection 10nM Mifepristone (InVitrogen), was added to the culture medium to induce the expression of the receptor. 18 hours post induction cells were detached from the flask using ethylenediamine tetra-acetic acid (EDTA; 1:5000; InVitrogen), following several washes with phosphate buffered saline pH 7.4 and resuspended in Sorting Medium containing Minimum Essential Medium (MEM), without phenol red, and supplemented with Earles salts and 3% Foetal Clone II (Hyclone). Approximately 1x 10e7 cells were examined for receptor expression by staining with a rabbit polyclonal antibody, 4a, raised against the N-terminal domain of the histamine H3 receptor, incubated on ice for 60 minutes, followed by two washes in sorting medium. Receptor bound antibody was detected by incubation of the cells for 60 minutes on ice with a goat anti rabbit antibody, conjugated with Alexa 488 fluorescence marker (Molecular Probes). Following two further washes with Sorting Medium, cells were filtered through a 50μm Filcon™ (BD Biosciences) and then analysed on a FACS Vantage SE Flow Cytometer fitted with an Automatic Cell Deposition Unit. Control cells were non-induced cells treated in a similar manner. Positively stained cells were sorted as single cells into 96-well plates, containing Complete Medium containing 500μg ml^{-1}

Zeocin™ and allowed to expand before reanalysis for receptor expression via antibody and ligand binding studies. One clone, 3H3, was selected for membrane preparation.

(ii) Membrane preparation from cultured cells

All steps of the protocol are carried out at 4°C and with pre-cooled reagents. The cell pellet is resuspended in 10 volumes of buffer A2 containing 50mM N-2-hydroxyethylpiperazine-N'-2-ethanesulfonic acid (HEPES) (pH 7.40) supplemented with 10e-4M leupeptin (acetyl-leucyl-leucyl-arginal; Sigma L2884), 25µg/ml bacitracin (Sigma B0125), 1mM ethylenediamine tetra-acetic acid (EDTA), 1mM phenylmethylsulfonyl fluoride (PMSF) and 2x10e-6M pepstain A (Sigma). The cells are then homogenised by 2 x 15 second bursts in a 1 litre glass Waring blender, followed by centrifugation at 500g for 20 minutes. The supernatant is then spun at 48,000g for 30 minutes. The pellet is resuspended in 4 volumes of buffer A2 by vortexing for 5 seconds, followed by homogenisation in a Dounce homogeniser (10-15 strokes). At this point the preparation is aliquoted into polypropylene tubes and stored at -70°C.

Compounds of the invention may be tested for in vitro biological activity in accordance with the following assays:

(I) Histamine H3 binding assay

For each compound being assayed, in a white walled clear bottom 96 well plate, is added:-

(a) 10µl of test compound (or 10µl of iodophenpropit (a known histamine H3 antagonist) at a final concentration of 10mM) diluted to the required concentration in 10% DMSO;

(b) 10µl ^{125}I 4-[3-(4-iodophenylmethoxy)propyl]-1H-imidazolium (iodoproxyfan) (Amersham; 1.85MBq/µl or 50µCi/ml; Specific Activity ~2000Ci/mmol) diluted to 200pM in assay buffer (50mM Tris(hydroxymethyl)aminomethane buffer (TRIS) pH 7.4, 0.5mM ethylenediamine tetra-acetic acid (EDTA)) to give 20pM final concentration; and

(c) 80µl bead/membrane mix prepared by suspending Scintillation Proximity Assay (SPA) bead type WGA-PVT at 100mg/ml in assay buffer followed by mixing with membrane (prepared in accordance with the methodology described above) and diluting in assay buffer to give a final volume of 80µl which contains 7.5µg protein and 0.25mg bead per well – mixture was pre-mixed at room temperature for 60 minutes on a roller.

The plate is shaken for 5 minutes and then allowed to stand at room temperature for 3-4 hours prior to reading in a Wallac Microbeta counter on a 1 minute normalised tritium count protocol. Data was analysed using a 4-parameter logistic equation.

(II) Histamine H3 functional antagonist assay

For each compound being assayed, in a white walled clear bottom 96 well plate, is added:-

(a) 10µl of test compound (or 10µl of guanosine 5'- triphosphate (GTP) (Sigma) as non-specific binding control) diluted to required concentration in assay buffer (20mM N-2-Hydroxyethylpiperazine-N'-2-ethanesulfonic acid (HEPES) + 100mM NaCl + 10mM $MgCl_2$, pH7.4 NaOH);

(b) 60µl bead/membrane/GDP mix prepared by suspending wheat germ agglutinin-polyvinyltoluene (WGA-PVT) scintillation proximity assay (SPA) beads at 100mg/ml in assay buffer followed by mixing with membrane (prepared in accordance with the methodology described above) and diluting in assay buffer to give a final volume of 60µl which contains 10µg protein and 0.5mg bead per well – mixture is pre-mixed at 4°C for 30 minutes on a roller and just prior to addition to the plate, 10µM final concentration of guanosine 5' diphosphate (GDP) (Sigma; diluted in assay buffer) is added;

The plate is incubated at room temperature to equilibrate antagonist with receptor/beads by shaking for 30 minutes followed by addition of:

(c) 10µl histamine (Tocris) at a final concentration of 0.3µM; and

(d) 20µl guanosine 5' [γ35-S] thiotriphosphate, triethylamine salt (Amersham; radioactivity concentration = 37kBq/µl or 1mCi/ml; Specific Activity 1160Ci/mmol) diluted to 1.9nM in assay buffer to give 0.38nM final.

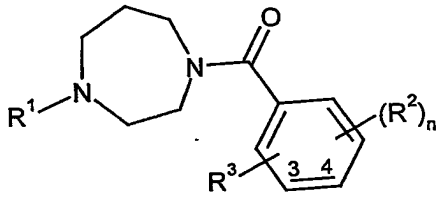
The plate is then incubated on a shaker at room temperature for 30 minutes followed by centrifugation for 5 minutes at 1500 rpm. The plate is read between 3 and 6 hours after completion of centrifuge run in a Wallac Microbeta counter on a 1 minute normalised tritium count protocol. Data is analysed using a 4-parameter logistic equation. Basal activity used as minimum i.e. histamine not added to well.

Results

The compounds of Examples E1-E10 were tested in the histamine H3 functional antagonist assay and exhibited pK_b values > 8.0. More particularly, the compounds of Examples 1-9 exhibited pK_b values > 9.0.

CLAIMS:

1. A compound of formula (I) or a pharmaceutically acceptable salt thereof:



(I)

wherein:
R^1 represents branched C_{3-6} alkyl, C_{3-5} cycloalkyl or -C_{1-4} alkylC_{3-4} cycloalkyl;
R^2 represents halogen, C_{1-6} alkyl, C_{1-6} alkoxy, cyano, amino or trifluoromethyl;
n represents 0, 1 or 2;
R^3 represents –X-aryl, -X-heteroaryl, -X-heterocyclyl, -X-aryl-aryl, -X-aryl-heteroaryl, -X-aryl-heterocyclyl, -X-heteroaryl-aryl, -X-heteroaryl-heteroaryl, -X-heteroaryl-heterocyclyl, -X-heterocyclyl-aryl, -X-heterocyclyl-heteroaryl or –X-heterocyclyl-heterocyclyl;
such that when R^3 represents –X-piperidinyl, -X-piperidinyl-aryl, -X-piperidinyl-heteroaryl or –X-piperidinyl-heterocyclyl said piperidinyl group is attached to X via a nitrogen atom;
wherein R^3 is attached to the phenyl group of formula (I) at the 3 or 4 position;
X represents a bond, O, CO, SO_2, CH_2O, OCH_2 or C_{1-6} alkyl;
wherein said aryl, heteroaryl or heterocyclyl groups of R^3 may be optionally substituted by one or more (eg. 1, 2 or 3) halogen, hydroxy, cyano, nitro, oxo, haloC_{1-6} alkyl, polyhaloC_{1-6} alkyl, haloC_{1-6} alkoxy, polyhaloC_{1-6} alkoxy, C_{1-6} alkyl, C_{1-6} alkoxy, arylC_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkoxyC_{1-6} alkyl, C_{3-7} cycloalkylC_{1-6} alkoxy, C_{3-7} cycloalkylcarbonyl, -COC_{1-6} alkyl, C_{1-6} alkoxycarbonyl, arylC_{1-6} alkyl, heteroarylC_{1-6} alkyl, heterocyclylC_{1-6} alkyl, C_{1-6} alkylsulfonyl, C_{1-6} alkylsulfinyl, C_{1-6} alkylsulfonyloxy, C_{1-6} alkylsulfonylC_{1-6} alkyl, arylsulfonyl, arylsulfonyloxy, arylsulfonylC_{1-6} alkyl, aryloxy, -CO-aryl, -CO-heterocyclyl, -CO-heteroaryl, C_{1-6} alkylsulfonamidoC_{1-6} alkyl, C_{1-6} alkylamidoC_{1-6} alkyl, arylsulfonamido, arylaminosulfonyl, arylsulfonamidoC_{1-6} alkyl, arylcarboxamidoC_{1-6} alkyl, aroylC_{1-6} alkyl, arylC_{1-6} alkanoyl, or a group $NR^{15}R^{16}$, -NR^{15}CO-aryl, -NR^{15}CO-heterocyclyl, -NR^{15}CO-heteroaryl, -$CONR^{15}R^{16}$, -$NR^{15}COR^{16}$, -$NR^{15}SO_2R^{16}$ or -$SO_2NR^{15}R^{16}$ groups, wherein R^{15} and R^{16} independently represent hydrogen or C_{1-6} alkyl;
or solvates thereof.

2. A compound according to claim 1 which is a compound of formula E1-E10 or a pharmaceutically acceptable salt thereof.

3. A pharmaceutical composition which comprises the compound of formula (I) as defined in claim 1 or claim 2 or a pharmaceutically acceptable salt thereof and a pharmaceutically acceptable carrier or excipient.

4. A compound as defined in claim 1 or claim 2 for use in therapy.

5. A compound as defined in claim 1 or claim 2 for use in the treatment of neurological diseases.

6. Use of a compound as defined in claim 1 or claim 2 in the manufacture of a medicament for the treatment of neurological diseases.

7. A method of treatment of neurological diseases which comprises administering to a host in need thereof an effective amount of a compound of formula (I) as defined in claim 1 or claim 2 or a pharmaceutically acceptable salt thereof.

8. A pharmaceutical composition for use in the treatment of neurological diseases which comprises the compound of formula (I) as defined in claim 1 or claim 2 or a pharmaceutically acceptable salt thereof and a pharmaceutically acceptable carrier.